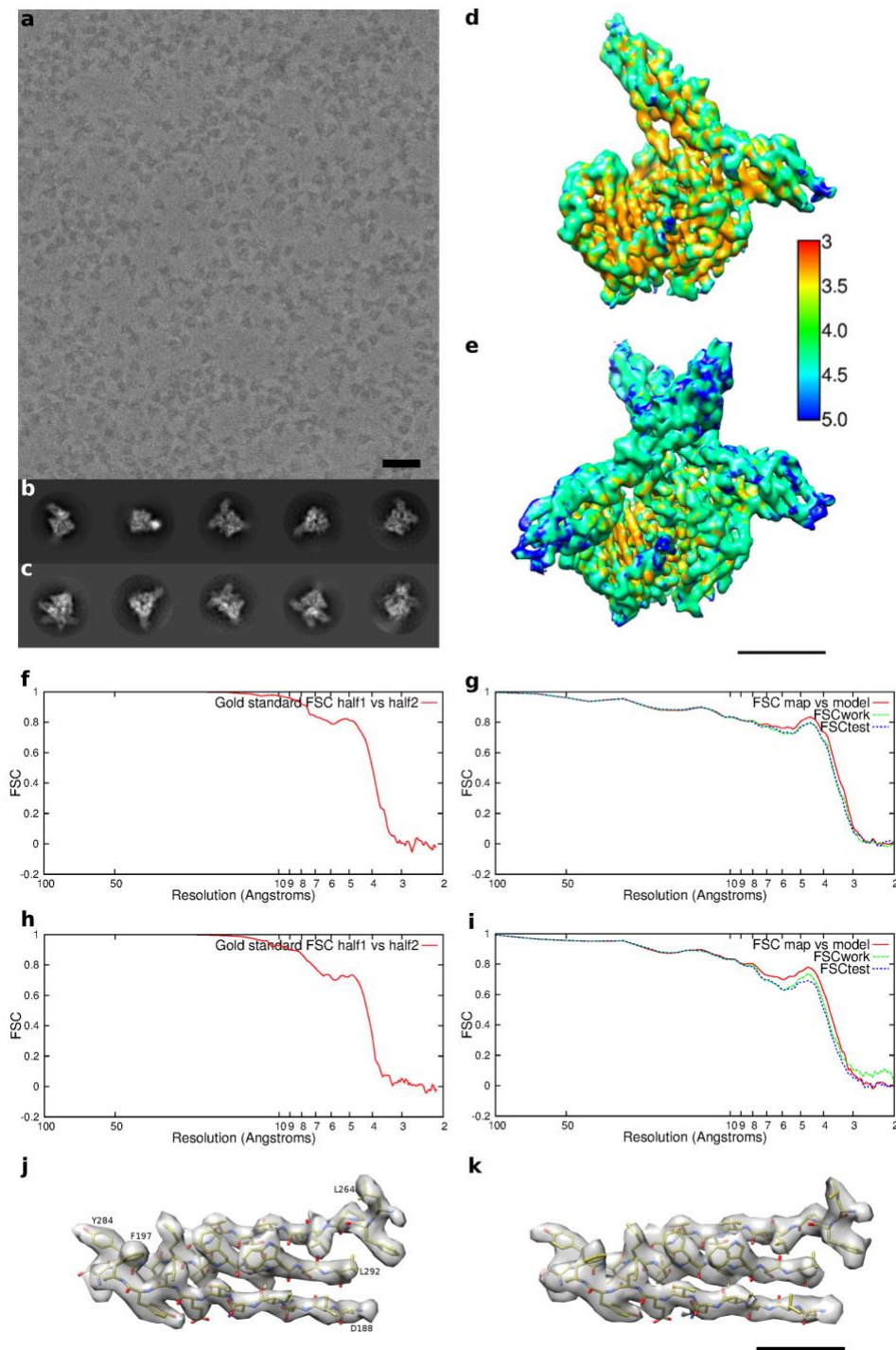


Cryo-EM structure of adenovirus type 3 fibre with desmoglein 2 shows an unusual mode of receptor engagement

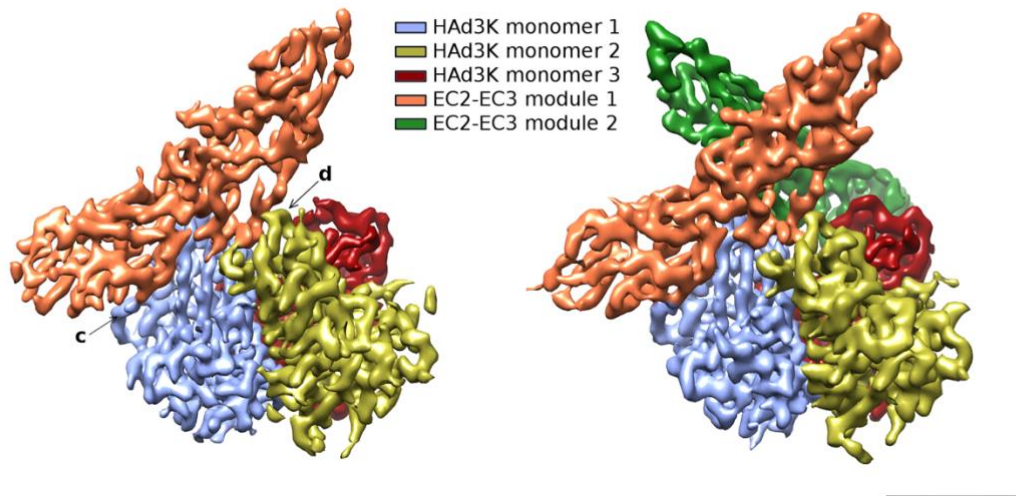
E. Vassal-Stermann, G. Effantin *et al.*,



Supplementary Figure 1 : **a-** Electron micrograph of HAd3K/EC2-EC3 complexes. **b & c-** 2D class averages for HAd3K in complex with one (**b**) or two (**c**) EC2-EC3 modules. **d & e-** 3D maps of HAd3K in complex with one (**d**) or two (**e**) EC2-EC3 modules colored according to local resolution. **f & h-** Fourier shell correlation (FSC) curves of the final 3D reconstructions obtained for HAd3K in complex with either one (**f**) or two (**h**) EC2-EC3 modules, the resolutions using FSC = 0.143 are 3.5 and 3.8 Å respectively. **g & i-** Cross-validation FSC curves for the refined model versus the summed map (FSC map vs model), half map 1 (FSCwork) and 2 (FSCtest) for HAd3K in complex with one (**g**) or two (**i**) DSG2 modules. **j & k-** Electron density of beta sheets fitted with the refined structure for the 3D reconstructions of HAd3K in complex with one (**j**) or two (**k**) DSG2 modules. Scale bars are 200 (Panel a), 30 (Panels d, e) and 10 Å (Panels j, k).

a - HAd3K / EC2-EC3

b - HAd3K / (EC2-EC3)₂



c Interface HAd3K monomer 1 / EC2 domain of module 1

d Interface HAd3K monomer 2 / EC3 domain of module 1



Supplementary Figure 2 : a, b - Isosurface views of the 3D reconstruction of HAd3K in complex with one (a) or two (b) EC2-EC3 modules. The subunits of both reconstructions are colored according to the color code. **c, d** – Close up views (arrows in (a)) for the 3D reconstruction of HAd3K in complex with one EC2-EC3 module. **c**- Zoom on residues at the interface of HAd3K monomer 1 and EC2 domain of EC2-EC3 module 1. **d**- Zoom on residues at the interface of HAd3K monomer 2 and EC3 domain of EC2-EC3 module1. Scale bars are 30 (Panels a, b) and 10 Å (Panels c, d). (*) Residue reported to reduce or abolish DSG2 binding in (Wang *et al.*, JVI 2013 : PMID 23946456)

Cryo-EM data collection

Magnification	130k
Pixel size (Å)	0.53
Defocus Range (μm)	-0.5 – -1.0
Voltage	300kV
Exposure dose	~1.5 e ⁻ /pixel/s
Exposure time (s per frame)	0.175
Number of frames	40
Total dose (e ⁻ /Å ²)	35

3D reconstruction summary

Box size (pixels)	200
Micrographs	2855
Autopicked particles	1799654
Particles after 2D classification	646863

	HAd3K with one DSG2	HAd3K with two
DSG2	module	modules
Particles included in the final 3D map	139958	78925
Resolution (Å) at FSC = 0.143	3.5	3.8
Model refinement statistics		
Resolution (Å) between atomic model and cryo-EM map	3.6 (FSC=0.5)	3.8 (FSC=0.5)
Ramachandran favored	92.0 %	90.0 %
Ramachandran outliers	0.0 %	0.0 %
Rotamer outliers	0.0 %	0.0 %
C-beta deviations	0	0
Clashscore	3.0	4.0
RMS(bonds)	0.0050	0.0051
RMS(angles)	1.17	1.20
MolProbity score	1.57	1.71

Supplementary Table 1: Data collection and refinement statistics

Residue Number	HAd3k/Ec2-Ec3	HAd3k/(Ec2-Ec3) ₂ Module 1	HAd3k/(Ec2-Ec3) ₂ Module 2
Ec2 LEU 122	✓	✓	✓
Ec2 ALA 124	✓	X	✓
Ec2 ALA 125	✓	✓	✓
Ec2 HIS 126	✓	✓	✓
Ec2 PRO 159	✓	✓	✓
Ec2 PRO 160	✓	✓	✓
Ec2 TYR 163	✓	✓	✓
Ec2 ASN 165	X	✓	✓
Ec2 LYS 166	X	✓	✓
Ec2 TYR 172	✓	✓	✓
Ec2 THR 173	✓	✓	X
Ec2 THR 174	✓ H	✓ H	✓
Ec2 SER 175	✓ H	✓ H	✓ H
Ec2 VAL 176	✓	✓	✓
Ec2 THR 177	✓ H	✓ H	✓ H
Ec2 LEU 178	✓	✓	✓
Ec2 ASP 179	✓	✓	✓ S
Ec2 GLU 182	✓	X	✓
Ec3 SER 314	✓	X	✓
Ec3 ILE 315	✓	X	X
Ec3 LYS 318	✓	X	✓
HAd3k ILE 145	✓	✓	✓
HAd3k GLU 146	X	X	✓
HAd3k TYR 147	✓	✓	✓
HAd3k GLY 148	✓	✓	✓
HAd3k GLN 150	X	✓	✓
HAd3k TYR 179	✓ H	✓ H	✓
HAd3k LEU 183	✓	✓	✓
HAd3k ASN 186 *	✓	✓	✓
HAd3k LYS 187	✓	✓	✓ S
HAd3k ASN 188	✓ H	✓ H	✓ H
HAd3k VAL 189 *	✓	✓	✓
HAd3k SER 190 *	✓ H	✓ H	✓ H
HAd3k ILE 191	✓	✓	✓
HAd3k ASN 192	✓	✓	✓
HAd3k GLU 268	X	X	✓
HAd3k LEU 289	✓	X	✓
HAd3k ASN 293	✓	✓	✓

Residues involved in the first interface of HAd3k with DSG2 identified by the PISA software
(Table 2, first page)

Residue Number	Had3k/Ec2-Ec3	Had3k/(Ec2-Ec3) ₂ Module 1	Had3k/(Ec2-Ec3) ₂ Module 2
Ec3 ALA 306	X	✓	✓
Ec3 PHE 311	✓	✓	✓
Ec3 HIS 312	X	✓	✓
Ec3 LYS 313	✓	✓	✓
Ec3 ARG 316	✓ H/S	✓ H/S	✓ H/S
Ec3 SER 317	✓ H	✓ H	✓ H
Ec3 TYR 319	✓	✓	✓
Ec3 LYS 320	✓	✓	✓ H
Ec3 PRO 321	✓	✓	✓
HAd3k LYS 187	✓	✓	✓
HAd3k ALA 259	✓	✓	✓
HAd3k ASP 261 *	✓ H/S	✓ H/S	✓ H/S
HAd3k GLY 262	✓	✓	✓
HAd3k ALA 263	✓	✓	✓ H
HAd3k LEU 264	✓ H	✓ H	✓ H
HAd3k PHE 265 *	✓	✓	✓
HAd3k PRO 266	✓	✓	✓
HAd3k ASN 293	✓	✓	✓
HAd3k ALA 294	✓	✓	✓
HAd3k GLY 295	✓	✓	✓
HAd3k LEU 296 *	✓	✓	✓
HAd3k ALA 297	✓ H	✓	✓
HAd3k PRO 298	✓	✓	✓ H
HAd3k GLU 299	X	X	✓

**Residues involved in the second interface of HAd3k with DSG2 identified by the PISA software
(Table2, second page)**

✓ Residue found at the interface

X Residue not found at the interface

H Residue found to form a putative hydrogen bound

S Residue found to form a putative salt bridge

* Residue reported to reduce or abolish DSG2 binding in (Wang *et al.*, JVI 2013 : PMID 23946456)

Supplementary Table 2: Interfacing residues between HAd3K and EC2 or EC3 domain of DSG2 identified by the PISA software

Primer Name	Primer Sequence
Ad3K-FWD-BamHI	TACATCGGATCCAGCTAATTCTATTGCACTTAAGAATAA
Ad3K-RVS-NotI	TACATCGCGGCCGCTCAGTCATCTTCTCTAATATAGGAAAAG
MBP- FWD-ATG-NdeI	TAGATCCATATGAAAATCGAAGAAGGTAAACTGG
EC3-RVS-KpnI	TAGATCGGTACCCTAAATGCCTTCTTTCACATTTTTCAC

Supplementary Table 3: Primers used for cloning in pET-Duet